EFSA CEF Panel (EFSA Panel on Food Contact Materials, Enzymes, Flavourings and Processing Aids), 2014. Scientific Opinion on Flavouring Group Evaluation 300, Revision 1 (FGE.300Rev1): One cyclo-aliphatic amide from chemical group 33

EFSA Publication

Link to article, DOI:
10.2903/j.efsa.2014.3887

Publication date:
2014

Document Version
Publisher's PDF, also known as Version of record

European Food Safety Authority

EFSA Journal 2014;12(11):3887

SCIENTIFIC OPINION

Scientific Opinion on Flavouring Group Evaluation 300, Revision 1 (FGE.300Rev1): One cyclo-aliphatic amide from chemical group 33[1]

EFSA Panel on Food Contact Materials, Enzymes, Flavourings and Processing Aids (CEF)[2, 3]

European Food Safety Authority (EFSA), Parma, Italy

ABSTRACT

The Panel on Food Contact Materials, Enzymes, Flavourings and Processing Aids of the European Food Safety Authority was requested to evaluate a flavouring substance, cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115] in the Flavouring Group Evaluation 300, Revision 1 (FGE.300Rev1) using the Procedure in Commission Regulation (EC) No 1565/2000. This revision is made due to a re-evaluation of the flavouring substance, cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115], as a 90-day dietary study in rats has become available. The substance was not considered to have genotoxic potential. The substance was evaluated through a stepwise approach (the Procedure) that integrates information on structure-activity relationships, intake from current uses, toxicological threshold of concern, and available data on metabolism and toxicity. The Panel concluded that the substance [FL-no: 16.115] does not give rise to safety concern at its levels of dietary intake estimated on the basis of the Maximised Survey-derived Daily Intake MSDI approach. Besides the safety assessment of this flavouring substance, the specifications for the material of commerce have also been considered. Specifications including complete purity criteria and identity for the material of commerce have been provided for the candidate substance.

KEY WORDS

flavouring, food safety, cyclo-aliphatic amide, FGE.300

1 On request from the Commission, Question No EFSA-Q-2014-00072, adopted on 23 October 2014.

2 Panel members: Claudia Bolognesi, Laurence Castle, Jean-Pierre Cravedi, Karl-Heinz Engel, Paul Fowler, Roland Franz, Konrad Grob, Rainer Gürtler, Trine Husøy, Wim Mennes, Maria Rosaria Milana, André Penninks, Maria de Fatima Tavares Poças, Vittorio Silano, Andrew Smith, Christina Tlustos, Fidel Toldra, Detlef Wölfle and Holger Zorn. Correspondence: fip@efsa.europa.eu

3 Acknowledgement: The Panel wishes to thank the members of the Working Groups on Flavourings: Ulla Beckman Sundh, Leon Brimer, Angelo Carere, Karl-Heinz Engel, Henrik Frandsen, Rainer Gürtler, Trine Husøy, Wim Mennes, Gerard Mulder and Harriet Wallin for the preparatory work on this scientific opinion and the hearing experts: Vibe Beltoft, Pia Lund and Karin Nørby and EFSA staff: Maria Carfí, Annamaria Rossi and Kim Rygaard Nielsen for the support provided to this scientific opinion.

Suggested citation: EFSA CEF Panel (EFSA Panel on Food Contact Materials, Enzymes, Flavourings and Processing Aids), 2014. Scientific Opinion on Flavouring Group Evaluation 300, Revision 1 (FGE.300Rev1): One cyclo-aliphatic amide from chemical group 33. EFSA Journal 2014;12(11):3887, 34 pp. doi:10.2903/j.efsa.2014.3887

Available online: www.efsa.europa.eu/efsajournal

SUMMARY

The European Food Safety Authority (EFSA) asked the Panel on Food Contact Materials, Enzymes, Flavourings and Processing Aids (the Panel) to deliver a scientific advice to the Commission on the implications for human health of chemically defined flavouring substances used in, or on, foodstuffs in the Member States. In particular, the Panel was requested to evaluate one flavouring substance in the Flavouring Group Evaluation 300, Revision 1 using the Procedure as referred to in the Commission Regulation (EC) No 1565/2000. The flavouring substance belongs to chemical group 30, Annex I of the Commission Regulation (EC) No 1565/2000.

The present Flavouring Group Evaluation deals with one cyclo-aliphatic amide. This revision is made due to additional toxicity data that have become available on the substance cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115].

The substance cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115] possesses three chiral centres and has been presented with specification of the stereoisomeric composition.

The substance is assigned into structural class III, according to the decision tree approach presented by Cramer et al., 1978.

The substance in the present group has not been reported to occur naturally in food.

In its evaluation, the Panel as a default used the "Maximised Survey-derived Daily Intake" (MSDI) approach to estimate the *per capita* intakes of the flavouring substances in Europe. However, when the Panel examined the information provided by the European Flavouring Industry on the use levels in various foods, it appeared obvious that the MSDI approach, in a number of cases, would grossly underestimate the intake by regular consumers of products flavoured at the use level reported by the Industry, especially in those cases where the annual production values were reported to be small. In consequence, the Panel had reservations about the data on use, and use levels, provided and the intake estimates obtained by the MSDI approach.

In the absence of more precise information that would enable the Panel to make a more realistic estimate of the intakes of the flavouring substances, the Panel has also decided to perform an estimate of the daily intakes per person using a "modified Theoretical Added Maximum Daily Intake" (mTAMDI) approach based on the normal use levels reported by Industry. In those cases where the mTAMDI approach indicated that the intake of a flavouring substance might exceed its corresponding threshold of concern, the Panel decided not to carry out a formal safety assessment using the Procedure. In these cases the Panel requires more precise data on use and use levels.

The results from the available limited genotoxicity studies do not raise a concern for genotoxicity and hence do not preclude the evaluation of the candidate substance in this FGE through the Procedure.

According to the default MSDI approach, the flavouring substance in this group has a total intake in Europe of 3 µg/*capita*/day, which is below the threshold of concern value for structural class III of 90 µg/person/day.

From the data available it is not possible to conclude that cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115] would be metabolised to innocuous products at the reported levels of intake as flavouring substance. Therefore, the substance was evaluated along the B-side of the Procedure. Now a 90-day study on the candidate substance [FL-no: 16.115] has become available and a No Observed Adverse Effect Level NOAEL to provide an adequate margin of safety of 2.9×10^6 is derived. Therefore, the substance [FL-no: 16.115] is not anticipated to pose a safety concern when used as flavouring substance at the estimated levels of intake, based on the MSDI approach.

When the estimated intake was based on the mTAMDI approach it was 960 µg/person/day for this flavouring substance belonging to structural class III. The estimated intake for the candidate substance is above the threshold of concern of 90 µg/person/day. Thus, for the flavouring substance considered in this opinion the intake, estimated on the basis of the mTAMDI, exceed the relevant threshold for the structural class, to which the flavouring substance has been assigned. Therefore, for the substance more reliable exposure data is required. On the basis of such additional data, the flavouring substance should be reconsidered along the steps of the Procedure. Following this procedure additional toxicological data might become necessary.

In order to determine whether the conclusion for the candidate substance can be applied to the material of commerce, it is necessary to consider the available specifications. Specifications including purity criteria and identity for the material of commerce have been provided for the flavouring substance.

In conclusion, for the flavouring substance cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115] the Panel considered that the substance would present no safety concern at the estimated level of intake estimated on the basis of the MSDI approach.

TABLE OF CONTENTS

BACKGROUND AS PROVIDED BY THE EUROPEAN COMMISSION

The use of flavourings is regulated under Regulation (EC) No 1334/2008 of the European Parliament and Council of 16 December 2008[4] on flavourings and certain food ingredients with flavouring properties for use in and on foods. On the basis of Article 9(a) of this Regulation, an evaluation and approval are required for flavouring substances.

The Union list of flavourings and source materials was established by Commission Implementing Regulation (EC) No 872/2012[5]. The list contains flavouring substances for which the scientific evaluation should be completed in accordance with Commission Regulation (EC) No 1565/2000[6].

EFSA has evaluated cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115] in the flavouring group evaluation 300 (FGE.300). The Opinion was adopted on 6 July 2011. EFSA concluded that for substance [FL-no: 16.115] no appropriate NOAEL was available and additional data were required.

The requested information on cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115] has now been submitted by the applicant.

The Commission asks EFSA to evaluate this new information and depending on the outcome proceed to the full evaluation of the flavouring substances.

TERMS OF REFERENCE AS PROVIDED BY THE EUROPEAN COMMISSION

The European Commission requests EFSA to carry out a safety assessment on the following flavouring substance: cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115] in accordance with Commission Regulation (EC) No 1565/2000.

[4] Regulation (EC) No 1334/2008 of the European Parliament and of the Council of 16 December 2008 on flavourings and certain food ingredients with flavouring properties for use in and on foods and amending Council Regulation (EEC) No 1601/91, Regulations (EC) No 2232/96 and (EC) No 110/2008 and Directive 2000/13/EC. OJ L 354, 31.12.2008, p. 34-50.

[5] Commission implementing Regulation (EU) No 872/2012 of 1 October 2012 adopting the list of flavouring substances provided for by Regulation (EC) No 2232/96 of the European Parliament and of the Council, introducing it in Annex I to Regulation (EC) No 1334/2008 of the European Parliament and of the Council and repealing Commission Regulation (EC) No 1565/2000 and Commission Decision 1999/217/EC. OJ L 267, 2.10.2012, p, 1-161.

[6] Commission Regulation No 1565/2000 of 18 July 2000 laying down the measures necessary for the adoption of an evaluation programme in application of Regulation (EC) No 2232/96. OJ L 180, 19.7.2000, p. 8-16.

ASSESSMENT

1. History of the Evaluation of the Substances in FGE.300Rev1

In FGE.300, the Panel evaluated cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115]. The Panel concluded that for the candidate substance [FL-no: 16.115], no appropriate NOAEL was available and additional data were required.

FGE	Opinion adopted	Link	No. of substances
FGE.300	6 July 2011	http://www.efsa.europa.eu/en/efsajournal/pub/2180.htm	1
FGE.300Rev1			1

The present revision of FGE.300, FGE.300Rev1, includes a re-evaluation of cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115], as additional data, a 14-day and a 90-day dietary study in rat have become available (Koetzner, 2013b; Koetzner, 2013a). No further data on the candidate substance [FL-no: 16.115] were found in the public literature.

A 90-day dietary study in rat on the supporting substance *N*-3,7-dimethyl-2,6-octadienyl cyclopropylcarboxamide [FL-no: 16.095] (Bauter, 2011) and additional information on stereoisomerism (Flavour Industry, 2013) have also been included in this revision of FGE.300.

2. Presentation of the Substances in Flavouring Group Evaluation 300, Revision 1

2.1. Description

The present Flavouring Group Evaluation 300, Revision 1 (FGE.300Rev1), using the Procedure as referred to in the Commission Regulation (EC) No 1565/2000 (EC, 2000) (The Procedure - shown in schematic form in Appendix A of this FGE), deals with one cyclo-aliphatic amide from chemical group 33, Annex I of Commission Regulation (EC) No 1565/2000 (EC, 2000). The flavouring substance, cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115] under consideration, as well as the chemical Register name, FLAVIS- (FL-), Chemical Abstract Service- (CAS-), Council of Europe- (CoE-) and Flavor and Extract Manufactures Association- (FEMA-) numbers, structure and specifications, are listed in Table 1.

The Panel is aware that there are several other amides in the Register considered in FGE.86 and FGE.94 and evaluated in FGE.304. For example, *N*-ethyl-2-isopropyl-5-methylcyclohexane carboxamide [FL-no: 16.013] from FGE.86, *N*1-(2-methoxy-4-methylbenzyl)-*N*2-(2-(pyridin-2-yl)ethyl)oxalamide [FL-no: 16.101], *N*-[(ethoxycarbonyl)methyl]-p-menthane-3-carboxamide [FL-no: 16.111] and cyclopropanecarboxamide, *N*-[(2E)-3,7-dimethyl-2,6-octadien-1-yl]- [FL-no: 16.095] from FGE.94, were evaluated by the JECFA and considered by the Panel. *N*-p-benzeneacetonitrile-menthanecarboxamide [FL-no: 16.117] and *N*-(2-(pyridine-2-yl)ethyl-3-p-menthanecarboxamide [FL-no: 16.118] were evaluated by the Panel in FGE.304. Of these *N*-3,7-dimethyl-2,6-octadienyl cyclopropylcarboxamide [FL-no: 16.095] shows the highest structural similarity to the candidate substance owing to the cyclopropancarboxylic acid moiety, and will as the only one of the amides in the Register be used to support the evaluation of the candidate substance in the present FGE.

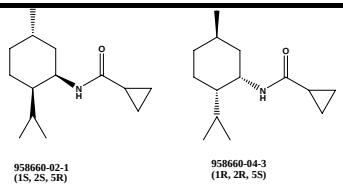
The outcome of the Safety Evaluation is summarised in Table 4.

The hydrolysis products of the candidate amide are listed in Table 5.

The supporting substance is listed in Table 6, together with its evaluation status.

SUMMARY OF SPECIFICATION DATA

Table 1: Specification Summary of the Substances in the Flavouring Group Evaluation 300Rev1

FL-no	EU Register name	Structural formula	FEMA no CoE no CAS no	Phys.form Mol.formula Mol.weight	Solubility [(a)] Solubility in ethanol [(b)]	Boiling point, °C [(c)] Melting point, °C ID test Assay minimum	Refrac. Index [(d)] Spec.gravity [(e)]	Specification comments
16.115	Cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide	 958660-02-1 (1S, 2S, 5R) 958660-04-3 (1R, 2R, 5S)	4558 958660-02-1	Solid $C_{14}H_{25}NO$ 223.35	Insoluble Soluble	 166 MS 98 %	n.a. n.a.	Two CASrn assigned 958660-02-1 (1S,2S,5*R*) and 958660-04-3 (1*R*,2*R*,5*S*). Min assay is sum of isomers: two main isomers (48 % of each) and 1 % of each of the two other stereoisomers (1*R*,2*R*,5*R*) and (1*S*,2*S*,5*S*) (Flavour Industry, 2013).

(a): Solubility in water, if not otherwise stated.
(b): Solubility in 95 % ethanol, if not otherwise stated.
(c): At 1013.25 hPa, if not otherwise stated.
(d): At 20°C, if not otherwise stated.
(e): At 25°C, if not otherwise stated.

2.2. Stereoisomers

It is recognised that geometrical and optical isomers of substances may have different properties. Their flavour may be different, they may have different chemical properties resulting in possible variability in their absorption, distribution, metabolism, elimination and toxicity. Thus, information must be provided on the configuration of the flavouring substance, i.e. whether it is one of the geometrical/optical isomers, or a defined mixture of stereoisomers. The available specifications of purity will be considered in order to determine whether the safety evaluation carried out for candidate substances for which stereoisomers may exist can be applied to the material of commerce. Flavouring substances with different configurations should have individual chemical names and codes (CAS number, FLAVIS number etc.).

The candidate substance cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115] possesses three chiral centres and has been presented with specification of the stereoisomeric composition (Flavour Industry, 2013).

2.3. Natural Occurrence in Food

The candidate substance [FL-no: 16.115] has not been reported to occur naturally in any food (TNO, 2009).

3. Specifications

Purity criteria for the substance have been provided by the Flavour Industry (Flavour Industry, 2009) (Table 1).

Judged against the requirements in Annex II of Commission Regulation (EC) No 1565/2000 (EC, 2000), this information is adequate for the candidate substance.

4. Intake Data

Annual production volumes of the flavouring substances as surveyed by the Industry can be used to calculate the "Maximised Survey-derived Daily Intake" (MSDI) by assuming that the production figure only represents 60 % of the use in food due to underreporting and that 10 % of the total EU population are consumers (SCF, 1999).

However, the Panel noted that due to year-to-year variability in production volumes, to uncertainties in the underreporting correction factor and to uncertainties in the percentage of consumers, the reliability of intake estimates on the basis of the MSDI approach is difficult to assess.

The Panel also noted that in contrast to the generally low per capita intake figures estimated on the basis of this MSDI approach, in some cases the regular consumption of products flavoured at use levels reported by the Flavour Industry in the submissions would result in much higher intakes. In such cases, the human exposure thresholds below which exposures are not considered to present a safety concern might be exceeded.

Considering that the MSDI model may underestimate the intake of flavouring substances by certain groups of consumers, the SCF recommended also taking into account the results of other intake assessments (SCF, 1999).

One of the alternatives is the "Theoretical Added Maximum Daily Intake" (TAMDI) approach, which is calculated on the basis of standard portions and upper use levels (SCF, 1995) for flavourable beverages and foods in general, with exceptional levels for particular foods. This method is regarded as a conservative estimate of the actual intake by most consumers because it is based on the assumption that the consumer regularly eats and drinks several food products containing the same flavouring substance at the upper use level.

One option to modify the TAMDI approach is to base the calculation on normal rather than upper use levels of the flavouring substances. This modified approach is less conservative (e.g., it may underestimate the intake of consumers being loyal to products flavoured at the maximum use levels reported) (EC, 2000). However, it is considered as a suitable tool to screen and prioritise the flavouring substances according to the need for refined intake data (EFSA, 2004).

4.1. Estimated Daily *per Capita* Intake (MSDI Approach)

The intake estimation is based on the Maximised Survey-derived Daily Intake (MSDI) approach, which involves the acquisition of data on the amounts used in food as flavourings (SCF, 1999). These data are derived from surveys on annual production volumes in Europe. These surveys were conducted in 1995 by the International Organization of the Flavour Industry, in which flavour manufacturers reported the total amount of each flavouring substance incorporated into food sold in the EU during the previous year (IOFI, 1995). The intake approach does not consider the possible natural occurrence in food.

Average per capita intake (MSDI) is estimated on the assumption that the amount added to food is consumed by 10 % of the population[7] (Eurostat, 1998). This is derived for candidate substances from estimates of annual volume of production provided by Industry and incorporates a correction factor of 0.6 to allow for incomplete reporting (60 %) in the Industry surveys (SCF, 1999).

The annual volume of production of the candidate substance in the present Flavouring Group Evaluation (FGE.300Rev1) from use as flavouring substance in Europe has been reported to be approximately 25 kg (Flavour Industry, 2009), and for the supporting substance to be approximately 500 kg (Flavour Industry, 2004). The daily *per capita* intakes for the candidate and the supporting substance are 3.0 and 61 µg, respectively (Table 4).

4.2. Intake Estimated on the Basis of the Modified TAMDI (mTAMDI)

The method for calculation of modified Theoretical Added Maximum Daily Intake (mTAMDI) values is based on the approach used by SCF up to 1995 (SCF, 1995).

The assumption is that a person may consume a certain amount of flavourable foods and beverages per day.

For the candidate substance information on food categories and normal and maximum use levels[8,9] were submitted by the Flavour Industry (Flavour Industry, 2009). The candidate substance is used in flavoured food products divided into the food categories, outlined in Annex III of the Commission Regulation (EC) No 1565/2000 (EC, 2000), as shown in Table 2. For the present calculation of mTAMDI, the reported normal use levels were used. In the case where different use levels were reported for different food categories the highest reported normal use level was used.

Table 2: Use of the Candidate Substance

Food category	Description	Flavouring used
01.0	Dairy products, excluding products of category 2	Yes
02.0	Fats and oils, and fat emulsions (type water-in-oil)	Yes

[7] EU figure 375 million. This figure relates to EU population at the time for which production data are available, and is consistent (comparable) with evaluations conducted prior to the enlargement of the EU. No production data are available for the enlarged EU.

[8] "Normal use" is defined as the average of reported usages and "maximum use" is defined as the 95th percentile of reported usages (EFFA, 2002)

[9] The normal and maximum use levels in different food categories (EC, 2000) have been extrapolated from figures derived from 12 model flavouring substances (EFFA, 2004).

Table 2: Use of the Candidate Substance

Food category	Description	Flavouring used
03.0	Edible ices, including sherbet and sorbet	No
04.1	Processed fruits	No
04.2	Processed vegetables (incl. mushrooms & fungi, roots & tubers, pulses and legumes), and nuts & seeds	Yes
05.0	Confectionery	No
06.0	Cereals and cereal products, incl. flours & starches from roots & tubers, pulses & legumes, excluding bakery	No
07.0	Bakery wares	No
08.0	Meat and meat products, including poultry and game	Yes
09.0	Fish and fish products, including molluscs, crustaceans and echinoderms	Yes
10.0	Eggs and egg products	Yes
11.0	Sweeteners, including honey	No
12.0	Salts, spices, soups, sauces, salads, protein products etc.	Yes
13.0	Foodstuffs intended for particular nutritional uses	No
14.1	Non-alcoholic ("soft") beverages, excl. dairy products	Yes
14.2	Alcoholic beverages, incl. alcohol-free and low-alcoholic counterparts	No
15.0	Ready-to-eat savouries	Yes
16.0	Composite foods (e.g. casseroles, meat pies, mincemeat) - foods that could not be placed in categories 1 – 15	No

According to the Flavour Industry the normal use levels for the candidate substance are in the range of 0.2 - 10 mg/kg food, and the maximum use levels are in the range of 1.7 - 20 mg/kg (Flavour Industry, 2009).

The mTAMDI value is 960 µg/person/day for the candidate substance from structural class III (see Section 7).

For detailed information on use levels and intake estimations based on the mTAMDI approach, see Section 7 and Appendix B.

5. Absorption, Distribution, Metabolism and Elimination

Specific information regarding absorption, distribution, metabolism and excretion is not available for the candidate substance.

Simple aliphatic amides, such as formamide, acetamide, propionamide, n-butyramide and *n*-valeramide were reported to undergo hydrolysis in rabbits after oral administration. The extent of hydrolysis increased with increasing chain-length and ranged from 28 to 97 % of the dose. Complete hydrolysis was reported for phenylacetamide in rabbits. For the aliphatic amides, increased hydrolysis was seen with increased chain-lengths following incubation with rabbit liver extracts and liver slices (Bray et al., 1949).

Aliphatic and aromatic amides are expected to be partly metabolised to polar metabolites which are eliminated in the urine or bile (James, 1974; Schwen, 1982). Hydrolysis of the amide bond has been reported as a metabolic pathway for the amides dihydrocapsaicin and piperine *in vivo* in rats (Kawada and Iwai, 1985; Bhat and Chandrasekhara, 1987).

Like other aliphatic and aromatic amides the candidate substance is anticipated to be absorbed from the gastrointestinal tract and at least partly hydrolysed. However, due to the lack of specific

information on hydrolysis and metabolism and given the limited knowledge on biotransformation of amides structurally related to [FL-no: 16.115] it cannot be anticipated that the candidate substance is metabolised to innocuous products.

For more detailed information, see Appendix C.

6. Application of the Procedure for the Safety Evaluation of Flavouring Substances

The application of the Procedure is based on intakes estimated on the basis of the MSDI approach. Where the mTAMDI approach indicates that the intake of a flavouring substance might exceed its corresponding threshold of concern, a formal safety assessment is not carried out using the Procedure. In these cases the Panel requires more precise data on use and use levels. For comparison of the intake estimations based on the MSDI approach and the mTAMDI approach, see Section 6.

For the safety evaluation of the candidate substance from chemical group 33, the Procedure as outlined in Appendix A was applied, based on the MSDI approach. The stepwise evaluation of the substance is summarised in Table 4.

Step 1

The candidate substance cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115] is classified according to the decision tree approach by Cramer et al. (Cramer et al., 1978) into structural class III.

Step 2

Step 2 requires consideration of the metabolism of the candidate substances. The candidate substance [FL-no: 16.115], cannot be anticipated to be metabolised to innocuous products and thus the evaluation proceeds via the B-side of the Procedure.

Step B3

The estimated daily *per capita* intake of the candidate substance [FL-no: 16.115] is 3.0 µg, which is below the threshold for its structural class of 90 µg/person/day (class III). Accordingly, the evaluation of the substance proceeds to step B4 of the Procedure.

Step B4

For the candidate substance cyclopropanecarboxylic acid (2-isopropyl-5- methylcyclohexyl)-amide [FL-no: 16.115], a NOAEL of 147 mg/kg bw/day from a multiple dose 90-day oral toxicity study in rats was reported (Koetzner, 2013a). The estimated daily *per capita* intake of 3 µg corresponds to 0.05 µg/kg bw/day. Thus, a margin of safety of 2.9×10^6 can be calculated**.** Therefore, the substance [FL-no: 16.115] is not anticipated to pose a safety concern when used as flavouring substance at the estimated levels of intake, based on the MSDI approach.

7. Comparison of the Intake Estimations Based on the MSDI Approach and the mTAMDI Approach

The estimated intake for the candidate substance [FL-no: 16.115] assigned to structural class III, based on the mTAMDI, is 960 µg/person/day, which is above the threshold of concern of 90 µg/person/day.

Thus, for the candidate substance [FL-no: 16.115] further information is required. This would include more reliable intake data and then, if required, additional toxicological data.

For comparison of the MSDI and mTAMDI values, see Table 3.

Table 3: Estimated Intakes Based on the MSDI Approach and the mTAMDI Approach

FL-no	EU Register name	MSDI (µg/*capita*/day)	mTAMDI (µg/person/day)	Structural class	Threshold of concern (µg/person/day)
16.115	Cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide	3	960	Class III	90

8. Considerations of Combined Intakes from Use as Flavouring Substances

Because of structural similarities of candidate and supporting substances, it can be anticipated that many of the flavourings are metabolised through the same metabolic pathways and that the metabolites may affect the same target organs. Further, in case of combined exposure to structurally related flavourings, the pathways could be overloaded. Therefore, combined intake should be considered. As flavourings not included in this Flavouring Group Evaluation may also be metabolised through the same pathways, the combined intake estimates presented here are only preliminary. Currently, the combined intake estimates are only based on MSDI exposure estimates, although it is recognised that this may lead to underestimation of exposure. After completion of all FGEs, this issue should be readdressed. The total estimated combined daily *per capita* intake of structurally related flavourings is estimated by summing the MSDI for individual substances.

The total estimated combined daily *per capita* intake of structurally related flavourings is estimated by summing the MSDI for individual substances.

The combined intake of the candidate and supporting substance, both from structural class III, in Europe is estimated to be 64 µg/*capita*/day (3 and 61 µg/*capita*/day, respectively). This value is below the threshold of concern for a structural class III substance of 90 µg/person/day.

9. Toxicity

9.1. Acute Toxicity

Data available for the candidate and supporting substance report that the oral LD_{50} value, in rats, was greater than 2000 mg/kg body weight (bw).

The acute toxicity data are summarised in Table 7.

9.2. Subacute, Subchronic, Chronic and Carcinogenicity Studies

14-day oral toxicity study with cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115]

A 14-day study was performed with cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115] (Koetzner, 2013b). The study was performed according to OECD Guideline (TG 407) under GLP. Four groups of adult Crl: Sprague-Dawley® CD® IGS rats (5/sex/group) were maintained on diets containing 0 (control), 490, 1400, or 4900 mg/kg of cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide for 14 days, calculated to provide an average daily intake of 0, 43, 125, or 447 or 0, 46, 133, or 420 mg/kg bw/day, for males and females, respectively.

Results from the stability and concentration analyses of the test diets indicate that cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide was stable during presentation and at the target concentrations in the diet for all intake levels. Stability was assessed on days 0, 4, 7 and 10 after preparation at all concentrations. Based on the overall stability, homogeneity and

concentration verification analysis, animals were considered to have received target dietary doses of cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide.

There were no changes in clinical observations, body weight gain, food consumption (males) food efficiency, or gross findings that were considered a result of cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide administration. Group 4 female food consumption was reduced to 90 % of control; this was statistically significant.

Under the conditions of this 14-day test and based on the toxicological endpoints evaluated, these results indicate that male and female rats are expected to tolerate a repeated high dose dietary exposure equal to 4900 mg/kg feed of cyclopropanecarboxylic acid (2-isopropyl-5-methylcyclohexyl)-amide (approximately 420 - 450 mg/kg bw/day) in a study of longer duration.

90-day oral toxicity study with cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115]

A 90-day study was performed with cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115] (Koetzner, 2013a). The study was done according to OECD Guideline (TG 408) under GLP. Four groups of adult Crl: Sprague-Dawley® CD® IGS rats (10/sex/group) were given the substance through the diet at concentrations set to target exposure levels of 0, 40, 150 and 375 mg/kg bw/day, respectively, when feeding the animals *ad libitum*. This resulted in average daily intake levels of 39, 147, and 367 mg/kg bw/day for males and 40, 147, and 371 mg/kg bw/day for females, respectively, over the course of the study (days 0 - 91).

The animals were examined by focal illumination and indirect ophthalmoscopy prior to initiation and again at the end of the study (day 86), observed for viability twice daily and for signs of gross toxicity and behavioural changes at least once daily during the study and weekly for a battery of detailed clinical observations. Urine and blood samples were collected on day 83 from all study animals for urinalysis, hematology and clinical chemistry determinations and additional blood samples were collected for coagulation assessments on day 92/93, prior to necropsy. Gross necropsies and histological evaluation of selected organs and tissues according to OECD were performed on all study animals.

There were no mortalities during the study and no clinical observations attributable to cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide administration. There were no changes in body weight, body weight gain, food consumption, or food efficiency in male rats attributable to the administration of the test substance. At the highest test dose, female rats had decreases in terminal body weight (10 %) and body weight gain throughout the test (30% decrease in mean daily body weight gain over the entire study duration), that were concurrent with decreases in food consumption (*ca.* 10 % throughout the test) and food efficiency (20 % over the duration of the study) that are considered a dose-dependent effect of test substance administration. Similar effects on female body weights, body weight gain, food consumption and food efficiency were observed at the mid-dose, but to a less degree (- 7 %, - 22 %, -9 % and -16 %; same sequence as for the high dose females). Except for the decreased food consumption, the deviations from the control values in the high dose female group were statistically significant at the end of the study. For the mid dose group statistically significantly reduced mean daily food consumption was observed during some but not all study weeks and at the end of the study, none of the parameters discussed here showed statistical significance in the mid-dose group.

Statistically significant differences in clinical chemistry parameters were measured on day 83 in treated male and female rats compared to the control groups. However the changes were generally small in magnitude and within the range of historical control values. Based on histological findings in the liver (hypertrophy, but no morphological evidence of hepatocellular damage), these changes considered non-adverse. Liver weight and liver-to-body/brain weight ratio increases were observed in male and female rats administered the highest dietary dose. All other changes were incidental and

within the range of historical control values. Test substance-related, dose-dependent microscopic findings occurred in the liver of males and females administered the two highest dietary doses, characterised by minimal or slight hepatocellular hypertrophy, which appeared as enlargement of the centrilobular hepatocytes. This finding was present in 8/10 males and females (M: 5 minimal, 3 slight, F: 8 minimal) at the highest test substance dose, and 4/10 mid-dose males and females (M: 2 slight, 2 minimal; F: 4 minimal), with no occurrences at the lowest dietary dose administered or in control animals. There was no morphological evidence of hepatocellular damage. All remaining microscopic findings were not test substance-related and most were typical for the age and strain of rats used in this study.

The Panel considered that in absence of any other histopathological change in the liver and in absence of changes in clinical chemistry parameters, indicative for liver damage, the occurrence of slight to minimal centrilobular hypertrophy and the increases in liver weight are not adverse. However, the statistically significant decreases in body weight gain and terminal body weights in the high dose female group are of concern, especially since they are connected to a statistically significant decrease in food efficiency, which indicates a change in physiology. Therefore the Panel decided that based on the body weight observation in the females of the high dose group, the NOAEL from this study is at the mid dose of 147 mg/kg bw/day.

Repeated dose toxicity data are summarised Table 8.

Toxicity data on the supporting substance [FL-no: 16.095] are presented in Table 8 (see also FGE.94 Revision 1 for details).

9.3. Developmental / Reproductive Toxicity Studies

No data have been submitted, and no data on the candidate substance [FL-no: 16.115] were found when the open literature was searched.

9.4. Genotoxicity Studies

In vitro data are available for both the candidate and the supporting substance.

Candidate substance [FL-no: 16.115]

No genotoxic potential was observed when cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115] was incubated with *Salmonella typhimurium* strains TA98, TA100, TA102, TA1535 and TA1537 with or without metabolic activation at concentrations up to 1000 µg/plate (10, 31.6, 100, 316 and 1000 µg/plate) in two separate experiments using the plate incorporation method and the preincubation method. The authors noted that in the plate incorporation method with and without metabolic activation, the 1000 µg/plate concentration of the candidate substance was cytotoxic to the bacteria (August, 2007).

Supporting substance [FL-no: 16.095]

N-3,7-Dimethyl-2,6-octadienyl cyclopropylcarboxamide [FL-no: 16.095] was tested in a bacterial reverse mutation test using *S. typhimurium* strains TA97a, TA98, TA100 and TA1535 (doses of 10, 50, 100, 500 and 1000 µg/plate), and *E. coli* strain WP2uvrA with and without metabolic activation (doses of 50, 100, 500, 1000 and 2000 µg/plate). It was concluded to be negative for the induction of mutagenicity (Next Century Incorporated, 2004).

The results from the available limited genotoxicity studies do not raise a concern for genotoxicity and hence do not preclude the evaluation of the candidate substance in this FGE through the Procedure.

Genotoxicity data are summarised in Table 9.

CONCLUSION

The present Flavouring Group Evaluation deals with one cyclo-aliphatic amide. This revision is made due to additional toxicity data on cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115] has become available.

The candidate substance cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115] possesses three chiral centres and has been presented with specification of the stereoisomeric composition.

The substance is assigned into structural class III, according to the decision tree approach presented by Cramer et al., 1978.

The substance in the present group has not been reported to occur naturally in food.

The results from the available limited genotoxicity studies do not raise a concern for genotoxicity and hence do not preclude the evaluation of the candidate substance in this FGE through the Procedure.

From the data available, it is not possible to conclude that the candidate substance in this group cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115] would be metabolised to innocuous products at the reported levels of intake as flavouring substance. Therefore, the substance was evaluated along the B-side of the Procedure. For the candidate substance [FL-no: 16.115], a 90-day study has become available and a NOAEL to provide an adequate margin of safety of 2.9×10^6 is derived. Therefore, the substance [FL-no: 16.115] is not anticipated to pose a safety concern when used as flavouring substance at the estimated levels of intake, based on the MSDI approach.

According to the default MSDI approach, the flavouring substance in this group has a total intake in Europe of 3 µg/*capita*/day, which is below the threshold of concern value for structural class III of 90 µg/person/day.

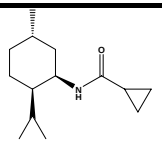
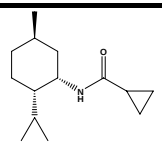
When the estimated intake was based on the mTAMDI approach it was 960 µg/person/day for this flavouring substance belonging to structural class III. The estimated intake for the candidate substance is above the threshold of concern of 90 µg/person/day. Thus, for the flavouring substance considered in this opinion the intake, estimated on the basis of the mTAMDI, exceed the relevant threshold for the structural class, to which the flavouring substance has been assigned. Therefore, for the substance more reliable exposure data is required. On the basis of such additional data, the flavouring substance should be reconsidered along the steps of the Procedure. Following this procedure additional toxicological data might become necessary.

In order to determine whether the conclusion for the candidate substance can be applied to the material of commerce, it is necessary to consider the available specifications. Specifications including purity criteria and identity for the material of commerce have been provided for the flavouring substance.

In conclusion, for the flavouring substance cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115] the Panel considered that the substance would present no safety concern at the estimated level of intake estimated on the basis of the MSDI approach.

SUMMARY OF SAFETY EVALUATION

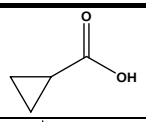
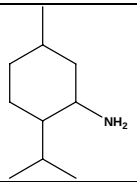
Table 4: Summary of Safety Evaluation Applying the Procedure

FL-no	EU Register name	Structural formula	MSDI [(a)] (µg/capita/day)	Class [(b)] Evaluation procedure path [(c)]	Outcome on the named compound [[(d)] or [(e)]]	Outcome on the material of commerce [[(f)], [(g)] or [(h)]]	Evaluation remarks
16.115	Cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide	 958660-02-1 (1S, 2S, 5R)  958660-04-3 (1R, 2R, 5S)	3	Class III B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	

(a): EU MSDI: Amount added to food as flavour in (kg / year) x 10^9 / (0.1 x population in Europe (= 375 x 10^6) x 0.6 x 365) = µg/*capita*/day.
(b): Thresholds of concern: Class I = 1800 µg/person/day, Class II = 540 µg/person/day, Class III = 90 µg/person/day.
(c): Procedure path A substances can be predicted to be metabolised to innocuous products. Procedure path B substances cannot.
(d): No safety concern based on intake calculated by the MSDI approach of the named compound.
(e): Data must be available on the substance or closely related substances to perform a safety evaluation.
(f): No safety concern at the estimated level of intake of the material of commerce meeting the specification requirement (based on intake calculated by the MSDI approach).
(g): Tentatively regarded as presenting no safety concern (based on intake calculated by the MSDI approach) pending further information on the purity of the material of commerce and/or information on stereoisomerism.
(h): No conclusion can be drawn due to lack of information on the purity of the material of commerce.

EVALUATION STATUS OF HYDROLYSIS PRODUCTS OF CANDIDATE SUBSTANCES

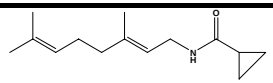
Table 5: Evaluation Status of Hydrolysis Products of Candidate Substances

FL-no	EU Register name JECFA no	Structural formula	SCF status [(a)] JECFA status [(b)] CoE status [(c)] EFSA status	Structural class [(d)] Procedure path (JECFA) [(e)]	Comments
Not in Reg.	Cyclopropanecarboxylic acid		Not evaluated as a flavour	- -	Not evaluated as a flavouring substance.
Not in Reg.	2-Isopropyl-5-methyl-cyclohexylamin		Not evaluated as a flavour	- -	Not evaluated as a flavouring substance.

(a): Category 1: Considered safe in use Category 2: Temporarily considered safe in use Category 3: Insufficient data to provide assurance of safety in use Category 4: Not acceptable due to evidence of toxicity.
(b): No safety concern at estimated levels of intake.
(c): Category A: Flavouring substance, which may be used in foodstuffs Category B: Flavouring substance which can be used provisionally in foodstuffs.
(d): Threshold of concern: Class I = 1800 µg/person/day, Class II = 540 µg/person/day, Class III = 90 µg/person/day.
(e): Procedure path A substances can be predicted to be metabolised to innocuous products. Procedure path B substances cannot.

SUPPORTING SUBSTANCE SUMMARY

Table 6: Supporting Substance Summary

FL-no	EU Register name	Structural formula	FEMA no CoE no CAS no	JECFA no Specification available	MSDI (EU) [(a)] (μg/*capita*/day)	SCF status [(b)] JECFA status [(c)] CoE status [(d)]	EFSA Comments
16.095	Cyclopropanecarboxamide, *N*-[(2E)-3,7-dimethyl-2,6-octadien-1-yl]-		4267 744251-93-2	1779	61	 No safety concern (JECFA, 2008)	

(a): EU MSDI: Amount added to food as flavouring substance in (kg / year) x 10E9 / (0.1 x population in Europe (= 375 x 10E6) x 0.6 x 365) = μg/capita/day.
(b): Category 1: Considered safe in use, Category 2: Temporarily considered safe in use, Category 3: Insufficient data to provide assurance of safety in use, Category 4: Not acceptable due to evidence of toxicity.
(c): No safety concern at estimated levels of intake.
(d): Category A: Flavouring substance, which may be used in foodstuffs, Category B: Flavouring substance which can be used provisionally in foodstuffs.

TOXICITY DATA

Table 7: Acute Toxicity

Chemical Name [FL-no][(a)]	Species	Sex	Route	LD_{50} (mg/kg bw)	Reference	Comments
Cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115]	Rat	F	Gavage	> 2000	(Vaeth, 2007)	
Cyclopropanecarboxamide, *N*-[(2E)-3,7-dimethyl-2,6-octadien-1-yl]- [FL-no: 16.095])	Rat	F	Gavage	>2000	(Merkel, 2004)	

F = Female.
(a): Supporting substances are listed in brackets

Table 8: Subacute / Subchronic / Chronic / Carcinogenicity Studies

Chemical Name [FL-no][(a)]	Species; Sex[2] No./Group[3]	Route	Dose levels	Duration	NOAEL (mg/kg bw/day)	Reference	Comments
(Cyclopropanecarboxamide, *N*-[(2E)-3,7-dimethyl-2,6-octadien-1-yl]- [FL-no: 16.095])	M; F 5/the two mid doses 10/control and high dose	Diet	0, 0.92, 9 and 92 mg/kg bw/day (M) 0, 0.98, 10 and 97 mg/kg bw/day (F)	28 days (unsupplemented feed for add. 14 days)	92	(Merkel, 2005)	OECD Guideline study (407)
	Rat; M, F 10		0, 0.7, 7.3, and 73.3 mg/kg bw/day (M) 0, 0.8, 8.1, and 80.1 mg/kg bw/day (F)	90-day	73	(Bauter, 2011)	OECD Guideline study (408).
Cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115]	Rat; M, F 5	Diet	0, 43, 125 and 447 mg/kg bw/day (M) 0, 46, 133 and 420 mg/kg bw/day (F)	14 days	approximately 420-447	(Koetzner, 2013b)	OECD Guideline study (407).
	Rat; M, F 10	Diet	39.2, 147.2, and 367.0 mg/kg bw/day (M) 39.6, 147.0, and 370.7 mg/kg bw/day (F)	90 days	147	(Koetzner, 2013a)	OECD Guideline study (408).

M = Male; F = Female.
(a): Supporting substances are listed in brackets.

Table 9: Genotoxicity Data (*in vitro*)

Chemical Name [FL-no][(a)]	Test System	Test Object	Concentration	Result	Reference	Comments
Cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115]	Ames test	*S. typhimurium* TA98; TA100; TA102; TA1535; TA1537	1000 µg/plate	Negative[(b)]	(August, 2007)	
(Cyclopropanecarboxamide, *N*-[(2E)-3,7-dimethyl-2,6-octadien-1-yl]- [FL-no: 16.095])	Ames test	*S. typhimurium* TA97a, TA98; TA100; TA1535 *E. Coli* WP2uvra	Up to 2000 µg/plate	Negative[(b)]	(Next Century Incorporated, 2004)	

(a): Supporting substances are listed in brackets.
(b): With and without S9 metabolic activation.

DOCUMENTATION PROVIDED TO EFSA

August M, 2007. Mutagenicity study of cyclopropancarbonsäure ((1*S*,2*S*,5*R*)-2-isopropyl-5-methyl-cyclohexyl)-amid, cyclopropancarbonsäure ((1*R*,2*R*,5*S*)-2-isopropyl-5-methylcyclohexyl)-amid in the *Salmonella typhimurium* reverse mutation assay (*in vitro*). LPT Report No. 18432/19/04. Laboratory of Pharmacology and Toxicology KG, Hamburg, Germany. Unpublished report submitted by EFFA to FLAVIS Secretariat.

Bauter MR, 2011. A repeated dose 90-day dietary study in rats. N-3,7-Dimethyl-2,6-octadienyl cyclopropylcarboxamide. Product Safety Labs. Study no. 31351. September 16, 2011. Unpublished report submitted by EFFA to FLAVIS Secretariat.

Flavour Industry, 2004. Unpublished information submitted by Flavour Industry to DG SANCO and forwarded to EFSA. A-94.

Flavour Industry, 2009. Unpublished information submitted by Flavour Industry to DG SANCO and forwarded to EFSA. A-300.

Flavour Industry, 2013. Unpublished information submitted by Flavour Industry to FLAVIS Secretariat. A-300 [FL-no: 16.115]. FLAVIS/8.226.

Koetzner L, 2013a. Cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide: a 90-day dietary study in rats. Product Safety Labs. Study no. 35939. December 2, 2013. Unpublished report submitted by EFFA to FLAVIS Secretariat.

Koetzner L, 2013b. Cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide: palatability/toxicity study: a 14-day dietary study in rats. Product Safety Labs. Study no. 35338. April 25, 2013. Unpublished report submitted by EFFA to FLAVIS Secretariat.

Merkel DJ, 2004. Acute oral toxicity up and down procedure in rats. Substance 04-237-01. Product Safety Laboratories. Study no. 16114. November 1, 2004. Unpublished report submitted by EFFA to Flavis Secretariat.

Merkel DJ, 2005. Subchronic toxicity study (28-day dietary study in rats). Product identification: 04-237-01. Product Safety Laboratories, Dayton, NJ, USA. Study No. 16600. June 30, 2005. Unpublished report submitted by EFFA to FLAVIS Secretariat.

Next Century Incorporated, 2004. Proprietary protection. 04-237: Bacterial reverse mutation test: plate incorporation and preincubation method for solids. Stirparo, B.S. Project no. 04-09-006. December 1, 2004. Unpublished report submitted by EFFA to FLAVIS Secretariat.

Vaeth A, 2007. Acute oral toxicity in the rat. Frey-Tox Lab No. 03015. Frey-Tox GmbH, Elster, Germany. Unpublished report submitted by EFFA to FLAVIS Secretariat.

REFERENCES

Bhat BG and Chandrasekhara N, 1986. Studies on the metabolism of piperine: absorption, tissue distribution and excretion of urinary conjugates in rats, Toxicology, 40, 83-92.

Bhat BG and Chandrasekhara N, 1987. Metabolic disposition of piperine in the rat. Toxicology 44, 99-106.

Bray HG, James SP, Thorpe WV, Wasdell MR and Wood PB, 1949. The fate of certain organic acids and amides in the rabbit. 9. Lower aliphatic amides. Biochemical Journal 45(4), 467-471.

Cramer GM, Ford RA and Hall RL, 1978. Estimation of toxic hazard - a decision tree approach. Food and Cosmetics Toxicology 16(3), 255-276.

EC (European Commission), 2000. Commission Regulation No 1565/2000 of 18 July 2000 laying down the measures necessary for the adoption of an evaluation programme in application of Regulation (EC) No 2232/96. Official Journal of the European Communities 19.7.2000, L 180, 8-16.

EFSA (European Food Safety Authority), 2004. Minutes of the 7th Plenary Meeting of the Scientific Panel on Food Additives, Flavourings, Processing Aids and Materials in Contact with Food, Held in Brussels on 12-13 July 2004. Brussels, 28 September 2004. [Online].

Eurostat, 1998. Total population. Cited in Eurostat, 2004. The EU population, Total population. December 2008. [Online].

IOFI (International Organization of the Flavor Industry), 1995. European inquiry on volume of use.

James R, 1974. Appendix 4, animal studies-summary of results. *N*-Ethyl-2-isopropyl-5-methylcyclohexane. Unpublished report to the Flavor Manufacturers Association. Submitted to WHO by the Flavor and Extract Manufacturers Association of the United States, Washington DC, USA.

JECFA (Joint FAO/WHO Expert Committee on Food Additives), 1995. Evaluation of certain food additives and contaminants. Forty-fourth Meeting of the Joint FAO/WHO Expert Committee on Food Additives. 14-23 February 1995. WHO Technical Report Series, no. 859. Geneva.

JECFA (Joint FAO/WHO Expert Committee on Food Additives), 1996. Toxicological evaluation of certain food additives. Forty-fourth Meeting of the Joint FAO/WHO Expert Committee on Food Additives and contaminants. WHO Food Additives Series: 35. IPCS, WHO, Geneva.

JECFA (Joint FAO/WHO Expert Committee on Food Additives), 1997. Evaluation of certain food additives and contaminants. Forty-sixth report of the Joint FAO/WHO Expert Committee on Food Additives. Geneva, 6-15 February 1996. WHO Technical Report Series, no. 868. Geneva.

JECFA (Joint FAO/WHO Expert Committee on Food Additives), 1999. Evaluation of certain food additives and contaminants. Forty-ninth report of the Joint FAO/WHO Expert Committee on Food Additives. Rome, 17-26 June 1997. WHO Technical Report Series, no. 884. Geneva.

JECFA (Joint FAO/WHO Expert Committee on Food Additives), 2006. Safety evaluation of certain food additives and contaminants. Sixty-third Meeting of the Joint FAO/WHO Expert Committee on Food Additives, WHO Food Additives Series: 54. IPCS, WHO, Geneva.

JECFA (Joint FAO/WHO Expert Committee on Food Additives), 2008. Safety evaluation of certain food additives and contaminants. Sixty-eight Meeting of the Joint FAO/WHO Expert Committee on Food Additives, WHO Food Additives Series: 59. IPCS, WHO, Geneva 2008. http://whqlibdoc.who.int/publications/2008/9789241660594_eng.pdf (May 2008).

Kawada T and Iwai K, 1985. *In vivo* and *in vitro* metabolism of dihydrocapsaicin, a pungent principle of hot pepper, in rats. Agricultural and Biological Chemistry 49 (2), 441-448.

SCF (Scientific Committee for Food), 1995. Scientific Committee for Food. First annual report on chemically defined flavouring substances. May 1995, 2nd draft prepared by the SCF Working Group on Flavouring Substances (Submitted by the SCF Secretariat, 17 May 1995). CS/FLAV/FL/140-Rev2. Annex 6 to Document III/5611/95, European Commission, Directorate-General III, Industry.

SCF (Scientific Committee for Food), 1999. Opinion on a programme for the evaluation of flavouring substances (expressed on 2 December 1999). Scientific Committee on Food. SCF/CS/FLAV/TASK/11 Final 6/12/1999. Annex I to the minutes of the 119th Plenary meeting. European Commission, Health & Consumer Protection Directorate-General.

Schwen R, 1982. Excretion and tissue distribution of ^{14}C after an oral dose of capsaicin analog, *N*-(vanillyl)-[1(14C)]-nonanamide. Federation Proceedings 41, 1558.

TNO (Nederlandse Organisatie voor Toegepast Natuurwetenschappelijk Onderzoek), 2009. VCF Volatile Compounds in Food. Nijssen LM, van Ingen-Visscher CA and Donders JJH (Eds.). Database version 11.1/11.11/12.1. Zeist, The Netherlands. TNO Triskelion, 1963-2009.

APPENDIX A: PROCEDURE FOR THE SAFETY EVALUATION

The approach for a safety evaluation of chemically defined flavouring substances as referred to in Commission Regulation (EC) No 1565/2000 (EC, 2000), named the "Procedure", is shown in schematic form in Figure A.1. The Procedure is based on the Opinion of the Scientific Committee on Food expressed on 2 December 1999 (SCF, 1999), which is derived from the evaluation Procedure developed by the Joint FAO/WHO Expert Committee on Food Additives at its 44th, 46th and 49th meetings (JECFA, 1995; JECFA, 1996; JECFA, 1997; JECFA, 1999).

The Procedure is a stepwise approach that integrates information on intake from current uses, structure-activity relationships, metabolism and, when needed, toxicity. One of the key elements in the Procedure is the subdivision of flavourings into three structural classes (I, II, III) for which thresholds of concern (human exposure thresholds) have been specified. Exposures below these thresholds are not considered to present a safety concern.

Class I contains flavourings that have simple chemical structures and efficient modes of metabolism, which would suggest a low order of oral toxicity. Class II contains flavourings that have structural features that are less innocuous, but are not suggestive of toxicity. Class III comprises flavourings that have structural features that permit no strong initial presumption of safety, or may even suggest significant toxicity (Cramer et al., 1978). The thresholds of concern for these structural classes of 1800, 540 or 90 µg/person/day, respectively, are derived from a large database containing data on subchronic and chronic animal studies (JECFA, 1996).

In Step 1 of the Procedure, the flavourings are assigned to one of the structural classes. The further steps address the following questions:

- can the flavourings be predicted to be metabolised to innocuous products[10] (Step 2)?
- do their exposures exceed the threshold of concern for the structural class (Step A3 and B3)?
- are the flavourings or their metabolites endogenous[11] (Step A4)?
- does a NOAEL exist on the flavourings or on structurally related substances (Step A5 and B4)?

In addition to the data provided for the flavouring substances to be evaluated (candidate substances), toxicological background information available for compounds structurally related to the candidate substances is considered (supporting substances), in order to assure that these data are consistent with the results obtained after application of the Procedure.

The Procedure is not to be applied to flavourings with existing unresolved problems of toxicity. Therefore, the right is reserved to use alternative approaches if data on specific flavourings warranted such actions.

[10] "Innocuous metabolic products": Products that are known or readily predicted to be harmless to humans at the estimated intakes of the flavouring agent" (JECFA, 1997).

[11] "Endogenous substances": Intermediary metabolites normally present in human tissues and fluids, whether free or conjugated; hormones and other substances with biochemical or physiological regulatory functions are not included (JECFA, 1997).

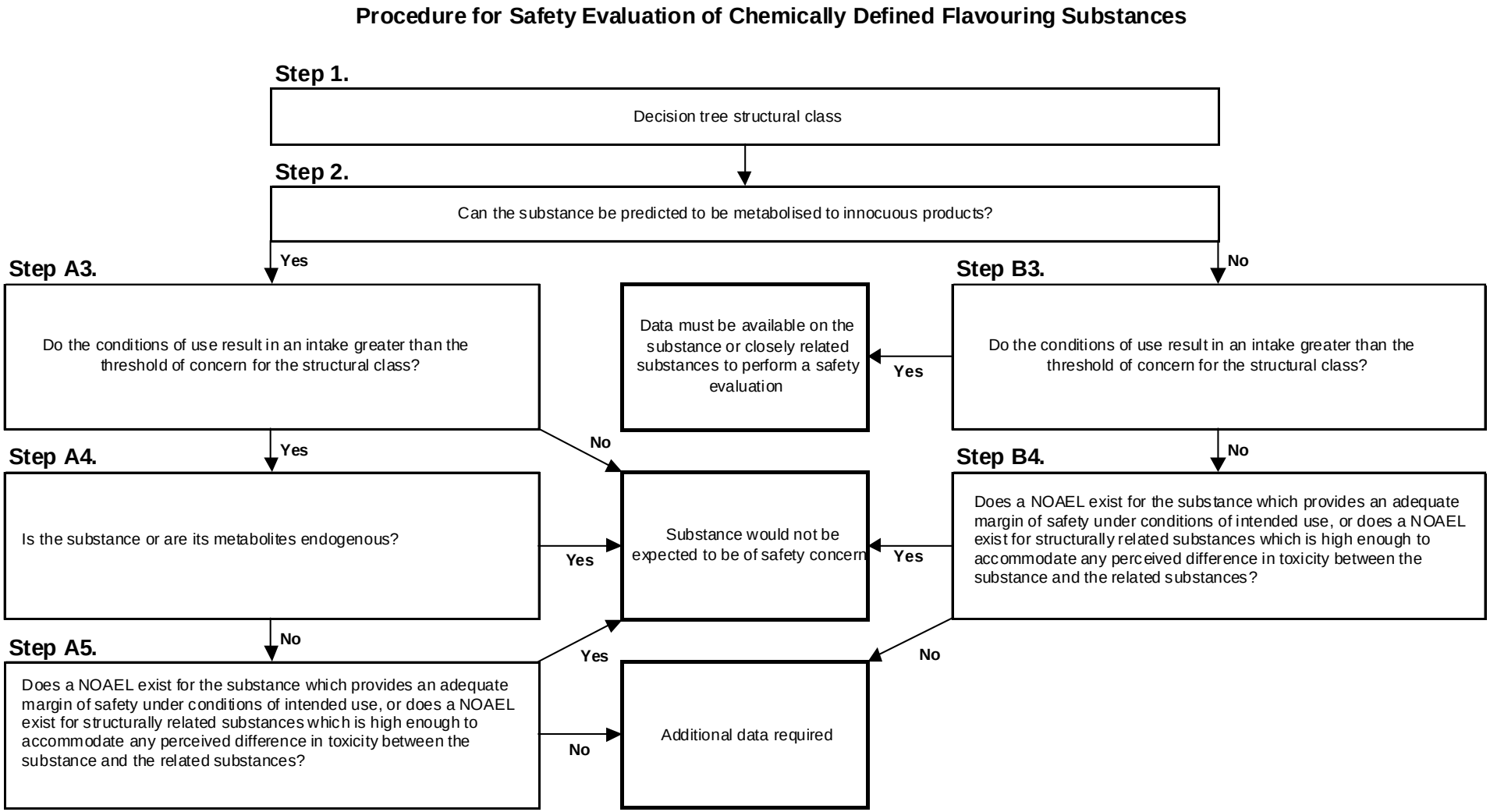


Figure A.1: Procedure for safety evaluation of chemically defined flavouring substances

APPENDIX B: USE LEVELS / mTAMDI

B.1 Normal and Maximum Use Levels

For each of the 18 Food categories (Table B.1.1) in which the candidate substances are used, Flavour Industry reports a "normal use level" and a "maximum use level" (EC, 2000). According to the Industry the "normal use" is defined as the average of reported usages and "maximum use" is defined as the 95th percentile of reported usages (EFFA, 2002). The normal and maximum use levels in different food categories have been extrapolated from figures derived from 12 model flavouring substances (EFFA, 2004).

Table B.1.1 Food categories according to Commission Regulation (EC) No 1565/2000 (EC, 2000)

Food category	Description
01.0	Dairy products, excluding products of category 02.0
02.0	Fats and oils, and fat emulsions (type water-in-oil)
03.0	Edible ices, including sherbet and sorbet
04.1	Processed fruit
04.2	Processed vegetables (incl. mushrooms & fungi, roots & tubers, pulses and legumes), and nuts & seeds
05.0	Confectionery
06.0	Cereals and cereal products, incl. flours & starches from roots & tubers, pulses & legumes, excluding bakery
07.0	Bakery wares
08.0	Meat and meat products, including poultry and game
09.0	Fish and fish products, including molluscs, crustaceans and echinoderms
10.0	Eggs and egg products
11.0	Sweeteners, including honey
12.0	Salts, spices, soups, sauces, salads, protein products, etc.
13.0	Foodstuffs intended for particular nutritional uses
14.1	Non-alcoholic ("soft") beverages, excl. dairy products
14.2	Alcoholic beverages, incl. alcohol-free and low-alcoholic counterparts
15.0	Ready-to-eat savouries
16.0	Composite foods (e.g. casseroles, meat pies, mincemeat) - foods that could not be placed in categories 01.0 - 15.0

The "normal and maximum use levels" are provided by Industry for the candidate substance in the present flavouring group (Table B.1.2).

Table B.1.2. Normal and Maximum use levels (mg/kg) for the candidate substances in FGE.300Rev1 (Flavour Industry, 2009).

FL-no	Food Categories																	
	Normal use levels (mg/kg) Maximum use levels (mg/kg)																	
	01.0	**02.0**	**03.0**	**04.1**	**04.2**	**05.0**	**06.0**	**07.0**	**08.0**	**09.0**	**10.0**	**11.0**	**12.0**	**13.0**	**14.1**	**14.2**	**15.0**	**16.0**
16.115	2 6	2 8	- -	- -	2 8	- -	- -	- -	2 8	2 8	4 10	- -	10 20	- -	0.2 1.7	- -	8 20	- -

B.2 mTAMDI Calculations

The method for calculation of modified Theoretical Added Maximum Daily Intake (mTAMDI) values is based on the approach used by SCF up to 1995 (SCF, 1995). The assumption is that a person may consume the amount of flavourable foods and beverages listed in Table B.2.1. These consumption estimates are then multiplied by the reported use levels in the different food categories and summed up.

Table B.2.1 Estimated amount of flavourable foods, beverages, and exceptions assumed to be consumed per person per day (SCF, 1995)

Class of product category	Intake estimate (g/day)
Beverages (non-alcoholic)	324.0
Foods	133.4
Exception a: Candy, confectionery	27.0
Exception b: Condiments, seasonings	20.0
Exception c: Alcoholic beverages	20.0
Exception d: Soups, savouries	20.0
Exception e: Others, e.g. chewing gum	e.g. 2.0 (chewing gum)

The mTAMDI calculations are based on the normal use levels reported by Industry. The seven food categories used in the SCF TAMDI approach (SCF, 1995) correspond to the 18 food categories as outlined in Commission Regulation (EC) No 1565/2000 (EC, 2000) and reported by the Flavour Industry in the following way (see Table B.2.2):

- Beverages (SCF, 1995) correspond to food category 14.1
- Foods (SCF, 1995) correspond to the food categories 1, 2, 3, 4.1, 4.2, 6, 7, 8, 9, 10, 13, and/or 16]
- Exception a (SCF, 1995) corresponds to food category 5 and 11
- Exception b (SCF, 1995) corresponds to food category 15
- Exception c (SCF, 1995) corresponds to food category 14.2
- Exception d (SCF, 1995) corresponds to food category 12

Exception e (SCF, 1995) corresponds to others, e.g. chewing gum.

Table B.2.2 Distribution of the 18 food categories listed in Commission Regulation (EC) No 1565/2000 (EC, 2000) into the seven SCF food categories used for TAMDI calculation (SCF, 1995)

	Food categories according to Commission Regulation (EC) No1565/2000	Distribution of the seven SCF food categories		
Key	Food category	Food	Beverages	Exceptions
01.0	Dairy products, excluding products of category 02.0	Food		
02.0	Fats and oils, and fat emulsions (type water-in-oil)	Food		
03.0	Edible ices, including sherbet and sorbet	Food		
04.1	Processed fruit	Food		
04.2	Processed vegetables (incl. mushrooms & fungi, roots & tubers, pulses and legumes), and nuts & seeds	Food		
05.0	Confectionery			Exception a
06.0	Cereals and cereal products, incl. flours &	Food		

Table B.2.2 Distribution of the 18 food categories listed in Commission Regulation (EC) No 1565/2000 (EC, 2000) into the seven SCF food categories used for TAMDI calculation (SCF, 1995)

	Food categories according to Commission Regulation (EC) No1565/2000	Distribution of the seven SCF food categories		
Key	Food category	Food	Beverages	Exceptions
	starches from roots & tubers, pulses & legumes, excluding bakery			
07.0	Bakery wares	Food		
08.0	Meat and meat products, including poultry and game	Food		
09.0	Fish and fish products, including molluscs, crustaceans and echinoderms	Food		
10.0	Eggs and egg products	Food		
11.0	Sweeteners, including honey			Exception a
12.0	Salts, spices, soups, sauces, salads, protein products, etc.			Exception d
13.0	Foodstuffs intended for particular nutritional uses	Food		
14.1	Non-alcoholic ("soft") beverages, excl. dairy products		Beverages	
14.2	Alcoholic beverages, incl. alcohol-free and low-alcoholic counterparts			Exception c
15.0	Ready-to-eat savouries			Exception b
16.0	Composite foods (e.g. casseroles, meat pies, mincemeat) - foods that could not be placed in categories 01.0 - 15.0	Food		

The mTAMDI value (see Table B.2.3) is presented for the flavouring substance in the present flavouring group (Flavour Industry, 2009). The mTAMDI value is only given for the highest reported normal use levels.

Table B.2.3 Estimated intakes based on the mTAMDI approach

FL-no	EU Register name	mTAMDI (µg/person/day)	Structural class	Threshold of concern (µg/person/day)
16.115	Cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide	960	Class III	90

APPENDIX C: METABOLISM

C.1. Introduction

The present FGE consists of one cyclo-aliphatic amide from chemical group 33: the candidate substance cyclopropanecarboxylic acid (2-isopropyl-5-methyl-cyclohexyl)-amide [FL-no: 16.115].

Specific information regarding absorption, distribution, metabolism and excretion is not available for the candidate substance.

C.2. Absorption, Distribution, Metabolism and Excretion (ADME)

JECFA (2006) in its evaluation of a group of aliphatic and aromatic amines and amides used as flavouring substances evaluated the ADME of a few amides.

The following text on absorption, distribution and elimination of amides is taken from the JECFA (JECFA, 2006):.

"Studies on selected members of the group indicate that amides per se are rapidly absorbed and metabolised".

"Male Sprague-Dawley rats given a single oral dose of 4 mg/kg bw of *N*-(vanillyl)-[1-^{14}C]nonamide (nonanyl 4-hydroxy-3-methoxybenzylamide) excreted 17.9 %, 45.9 % and 22.7 % of the radiolabel in the urine, faeces and expired CO_2, respectively, within 72 hours, although most of the radiolabel was excreted within the first 24 hours. Bile duct-cannulated rats excreted 11.4 %, 3.7 %, 11.7 % and 65.1 % of the radiolabel in the urine, faeces, expired CO_2 and bile, respectively. In fasted rats, peak blood levels of radiolabel occurred 10 minutes after administration. By 72 hours after dosing, the highest concentration of radiolabel was found in fat, liver and adrenal gland. These results indicate that nonanyl 4-hydroxy-3-methoxybenzylamide is rapidly absorbed and that appreciable quantities undergo enterohepatic circulation and partial conversion to CO_2 (Schwen, 1982)".

"Groups of male albino Wistar rats were given piperine at a dose of 170 mg/kg bw by gavage or 85 mg/kg bw by intraperitoneal injection, and urine and faeces were collected every 24 hours for 12 days. Urine and faeces from rats fed a control diet for 10 days were collected for 3 days before treatment and used as control samples. When given by either route, about 3 % of the unchanged dose was detected in faeces over 5 days, indicating that 97 % of the piperine was absorbed. Peak excretion in the faeces occurred on day 1 after intraperitoneal injection and on 3 days after gavage. No unchanged piperine was detected in urine after administration by either route; however, there was increased excretion of conjugated glucuronides, sulphates and phenols, with maxima on days 1-4. Overall, 91-97 % of the administered dose was accounted for. After treatment the animals were killed at various intervals, when blood was collected from the heart, and the liver, kidney, spleen and gut (stomach, small intestine, caecum and large intestine) were removed. By 30 minutes after ingestion of piperine, 29 % was detected in the gut (22 % in stomach and 6 % in small intestine). By 48 hours, 1 % was detected in stomach, and 2-3 % in the caecum and large intestine, indicating that 97 % had been absorbed. A similar pattern was reported in rats intraperitoneally injected with piperine, although some of the values differed (data not reported). Between 1 and 10 hours after treatment, only traces of piperine administered by either route were detected in blood. Between 0.5 and 24 hours after treatment, intraperitoneally administered piperine was detected in the liver (2.12-0.4 %) and kidney (0.04-0.2 %). Similarly, orally administered piperine was detected in the liver (0.25-0.12 %) and kidney (0.03-0.17 %) up to 24 hours after treatment. No piperine was detected after 48 hours in any of the tissues examined (Bhat and Chandrasekhara, 1986)".

"A group of male albino Wistar rats were given 175 mg/kg bw piperine by gavage. After 1 hour, some of the rats, including a group of untreated rats that served as controls, received a bile duct cannula, and bile was collected for 6 hours. Urine was collected from the remaining rats for 4 days and pooled, while urine collected for 4 days before dosing served as control samples. No unchanged piperine was detected in urine. Piperine was detected in the bile (about 1 % of the original dose) within 6 hours, and various metabolites (piperonylic acid, vanillic acid and piperonyl alcohol) were excreted in urine (about 15.5 % of the original dose) within 96 hours (Bhat and Chandrasekhara, 1987)".

"In rats given a single oral dose (not specified) of *N*-ethyl-para-menthane-[3-^{14}C]-carboxamide (*N*-ethyl-2-isopropyl-5-methylcyclohexanecarboxamide), 64.2 % and 28.7 % of the dose was excreted in urine and faeces, respectively, over 5 days. Almost 50 % of the radioactivity was secreted into the bile within 2 days, most within 24 hours, indicating enterohepatic circulation of the parent compound or its metabolites. The peak plasma concentration (0.3 % of the total dose) was reached within 1 hour. Subsequently, the compound was eliminated with a half-life of 11 hours. Whole-body autoradiography showed that most of the radioactivity was in the liver, kidneys and gastrointestinal tract. The results indicate that the substance was rapidly and extensively converted into more polar metabolites of unknown structure (James, 1974)".

The following text on metabolism, including hydrolysis, of amides is taken from JECFA (JECFA, 2006):

"The metabolic fate of *N*-ethyl-para-menthane-[3-^{14}C]-carboxamide was examined in one male and one female dog given a single dose oral dose of 10 mg/kg bw. The substance was readily absorbed end rapidly eliminated in the urine (72 % of the dose within the first 24 hours) and faeces (11 % of the dose within 5 days). No parent compound was detected in the urine. The main urinary metabolites were glucuronide or sulphate conjugates, whereas the faeces contained mainly unchanged compound. Radioactivity was detected (detection limit= 0.05 ppm) in the liver, adrenal glands (male only), testes and kidney (female only) 5 days after treatment. Peak plasma levels were reached within 4 hours. Subsequently the compound was eliminated with a half-life of about 70 minutes. Plasma radioactivity was determined to consist mostly (> 90 %) of metabolites of the test substance. About 70 % was bound to plasma protein in *vitro*, but < 10 % of the radioactivity was protein-bound in *vivo*. The author noted that rats metabolised the test substance to polar unconjugated metabolites, while dogs metabolised it to conjugates; however, both species metabolised it extensively and eliminated it rapidly (James, 1974)".

"These studies indicate that the amides in the group of flavouring agents are quickly absorbed, metabolised and excreted, mainly in urine but also partly in the faeces".

"Aliphatic amides have been reported to undergo limited hydrolysis. Extensive hydrolysis of aliphatic amides of various lengths was observed after incubation with rabbit liver extracts; however, hydrolysis was significantly slower for aliphatic amides with fewer than five or more than 10 carbons (Bray et al., 1949)".

"After administration of 1.5-5.0 g of acetamide or butyramide to rabbits, 62 % of the dose of acetamide was recovered unchanged in the urine within 24 hours, while only 13 % of the butyramide dose was recovered unchanged".

"Studies in which rats were given an oral dose (170 mg/kg bw) of piperine or dogs were given an oral dose (10 mg/kg bw) of *N*-ethyl-2-isopropyl-5-methylcyclohexanecarboxamide indicated that amide hydrolysis products are not major metabolites of these compounds (James, 1974; Bhat and Chandrasekhara, 1986)".

"The metabolism of piperine was studied in groups of male albino Wistar rats given a dose of 170 mg/kg bw by gavage or 85 mg/kg bw by intraperitoneal injection. Urine and faeces were collected every 24 hours for 12 days, while control urine and faeces samples were collected for 3 days from rats

fed a control diet before dosing. No unchanged piperine was detected in urine after exposure by either route; however, there was increased excretion of conjugated glucuronides, sulphates and phenols, with maximum excretion of all three on days 1 - 4. Demethylation of piperine was suggested by an increase in conjugated phenols. Over 8 days, about 36 % of the gavage dose was excreted in urine as conjugated phenols and 62 % as methylenedioxyphenyl metabolites. About 19 % of the intraperitoneal dose was excreted as phenolics and about 72 % as methylenedioxyphenyl derivatives (Bhat and Chandrasekhara, 1986). The proposed pathways for the metabolism of piperine in rats involved in addition of amide hydrolysis to piperic acid, metabolic oxidative cleavage of the benzylic alkene function results in a series of vanilloyl and piperonyl derivatives, which are excreted free of in conjugated form, mainly in the urine (Bhat and Chandrasekhara, 1987)".

The Panel, in addition to the studies identified by the JECFA, also retrieved the following study by (Bray et al., 1949; Kawada and Iwai, 1985) in which the metabolism in rats of dihydrocapsaicin was investigated *in vivo* and *in vitro*: Within 48 hours after oral administration of dihydrocapsaicin (20 mg/kg bw) to male adult rats, unchanged dihydrocapsaicin and eight of its metabolites were identified in urine; i.e. dihydrocapsaicin (8.7 % of total dose), vanillylamine (4.7 %), vanillin (4.6 %), vanillyl alcohol (37.6 %) and vanillic acid (19.2 %) as free forms and/or their glucuronides. The proportions of free and glucuronide metabolites in urine 14.5 % and 60.5 % of the total dose. Cell-free extracts of rat liver catalysed the hydrolysis of dihydrocapsaicin to vanillylamine and 8-methyl nonanoic acid. The former compound was further transformed to vanillin in situ. Dihydrocapsaicin-hydrolyzing enzyme activity was found in various organs of rats. The activity was located mainly in the liver (Kawada and Iwai, 1985).

The Panel also had a further look at the study performed by Bray et al. (1949) and concluded that simple aliphatic amides, such as formamide, acetamide, propionamide, n-butyramide, and n-valeramide were reported to undergo hydrolysis in rabbits after oral administration. The extent of hydrolysis increased with increasing chain-length and ranged from 28 to 97 % of the dose. Complete hydrolysis was reported for phenylacetamide in rabbits. For the aliphatic amides, increased hydrolysis was seen with increased chain-lengths following incubation with rabbit liver extracts and liver slices (Bray et al., 1949).

C.3. Summary and Conclusions

Specific information regarding absorption, distribution, metabolism and excretion is not available for the candidate substance.

Simple aliphatic amides, such as formamide, acetamide, propionamide, n-butyramide and n-valeramide were reported to undergo hydrolysis in rabbits after oral administration. The extent of hydrolysis increased with increasing chain-length and ranged from 28 to 97 % of the dose. Complete hydrolysis was reported for phenylacetamide in rabbits. For the aliphatic amides, increased hydrolysis was seen with increased chain-lengths following incubation with rabbit liver extracts and liver slices (Bray et al., 1949).

Aliphatic and aromatic amides are expected to be readily absorbed and partly metabolised to polar metabolites, which are eliminated in the urine or bile (James, 1974; Schwen, 1982). Hydrolysis of the amide bond has been reported as a metabolic pathway for the amides dihydrocapsaicin and piperine *in vivo* in rats (Bhat and Chandrasekhara, 1987).

In summary, like other aliphatic and aromatic amides, the candidate substance is anticipated to be absorbed from the gastrointestinal tract and at least partly hydrolysed. However, due to the lack of specific information on hydrolysis and metabolism and given the limited knowledge on biotransformation of amides structurally related to [FL-no: 16.115] it cannot be anticipated that the candidate substance is metabolised to innocuous products.

ABBREVIATIONS

ADI	Acceptable Daily Intake
ADME	Absorption, Distribution, Metabolism and Excretion
BW	Body weight
CAS	Chemical Abstract Service
CEF	Panel on Food Contact Materials, Enzymes, Flavourings and Processing Aids Chemical Abstract Service
CHO	Chinese hamster ovary (cells)
CoE	Council of Europe
DNA	Deoxyribonucleic acid
EC	European Commission
EFSA	The European Food Safety Authority
EU	European Union
FAO	Food and Agriculture Organization of the United Nations
FEMA	Flavor and Extract Manufacturers Association
FGE	Flavouring Group Evaluation
FLAVIS (FL)	Flavour Information System (database)
ID	Identity
IOFI	International Organization of the Flavour Industry
IR	Infrared spectroscopy
JECFA	The Joint FAO/WHO Expert Committee on Food Additives
LD_{50}	Lethal Dose, 50 %; Median lethal dose
MS	Mass spectrometry
MSDI	Maximised Survey-derived Daily Intake
mTAMDI	Modified Theoretical Added Maximum Daily Intake
NAD	Nicotinamide Adenine Dinucleotide
NADP	Nicotinamide Adenine Dinucleotide Phosphate
No	Number
NOAEL	No Observed Adverse Effect Level
NTP	National Toxicology Program
OECD	Organisation for Economic Co-operation and Development
SCE	Sister Chromatid Exchange
SCF	Scientific Committee on Food
SMART	Somatic Mutation and Recombination Test
TAMDI	Theoretical Added Maximum Daily Intake
UDS	Unscheduled DNA Synthesis
WHO	World Health Organisation